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DEVELOPMENT OF THE SUGARCANE BORER ON VARIOUS ARTIFICIAL DIETS

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DEVELOPMENT OF THE SUGARCANE BORER ON VARIOUS ARTIFICIAL DIETS

By F. D. Brewer[1]

ABSTRACT

In comparisons with a casein and wheat germ diet, a low agar, soybean, and wheat germ diet, and a pinto bean diet, a soyflour and wheat germ diet has these advantages for rearing the sugarcane borer, *Diatraea saccharalis* (F.): a shorter larval development time (20.8 days), a low larval mortality (4.4%), large pupae (males, 103.6 mg; females, 176.8 mg), and low cost. When the amounts of sugar and wheat germ were increased or alphacel was added to the low agar, soybean, and wheat germ diet (agar reduced by 80%), the pupal weights increased significantly. Shorter larval (20.9 days) and pupal (6.8 days) developmental periods were observed in borers reared on the soyflour and wheat germ diet when the agar content was reduced by 50% and corncob grits were substituted for the agar. Additional soyflour (25%) in the diet did not appear to be as important for shorter larval and pupal development time as did a reduction of the agar. **KEY WORDS: Artificial diets, development of *Diatraea saccharalis, Diatraea saccharalis,* insect nutrition, nutrition of *Diatraea saccharalis.***

INTRODUCTION

The rearing of host or pest species in the laboratory for use in biological control programs is common at many Agricultural Research Service stations. Two major considerations in the production of these species are the type and the cost of an artificial rearing medium. A research goal at the Bioenvironmental Insect Control Laboratory is the development of a diet that is both economical and nutritionally adequate to produce large numbers of healthy sugarcane borers, *Diatraea saccharalis* (F.)[2] (fig. 1).

Of the diets developed for rearing the sugarcane borer, a pinto bean diet (*3*)[3] and a casein and wheat germ diet (*4*) are two of the most widely used diets at the present time. Sanford (*5*) selected the pinto bean diet in place of the casein and wheat germ diet to rear the sugarcane borer because of a shorter larval developmental period and lower cost (about 50%).

Casein and agar are the most expensive ingredients in the casein and wheat germ diet. Shaver and Raulston (*6*) formulated an economical low agar soybean and wheat germ diet for rearing the tobacco budworm, *Heliothis virescens* (F.), by substituting a toasted defatted soyflour (Nutrisoy) for casein and by replacing 80% of the agar with corncob grits. Since soyflour was found to be a favorable substitute for the highly purified casein, the quality of protein should perhaps be given more emphasis than its quantity.

The purpose of this investigation was to determine the most suitable economical diet for rearing the sugarcane borer by comparing presently used diets, the low agar, soybean, and wheat germ diet, and a diet formulated in our laboratory. The replacement of casein and most of the agar with substitutes received special attention in regard to diet cost.

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[2] Lepidoptera: Pyralidae.

[3] Italic numbers in parentheses refer to items in "Literature Cited" at the end of this publication.

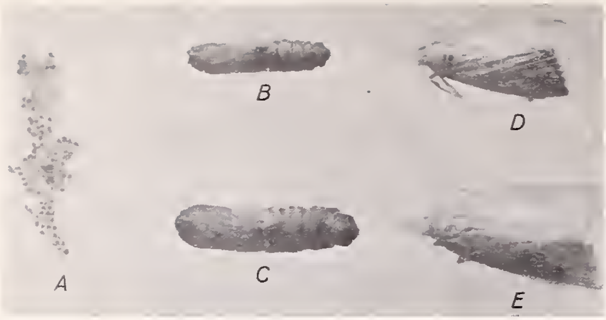

Figure 1.—Nonlarval stages of the sugarcane borer. *A*, Egg mass; *B*, male pupa (ventral view); *C*, female pupa (ventral view); *D*, male moth (dorsal view); and *E*, female moth (dorsal view).

MATERIALS AND METHODS

Direct replacement (gram for gram) of casein (minimum 90% protein) with Nutrisoy (minimum 50% protein) was made in the diet formulated by Berger (*2*), and then this diet was compared with the above three diets to determine developmental characteristics and diet costs. In a second experiment I compared modifications of this soyflour and wheat germ diet and the low agar, soybean, and wheat germ diet.

In our first experiment, the following four diets were compared: (A) soyflour and wheat germ (casein replaced with Nutrisoy as in Berger's diet); (B) pinto bean; (C) casein and wheat germ; and (D) low agar, soybean, and wheat germ. (See tables 1–4 for diet recipes.)

Table 1.—*Composition of a soyflour and wheat germ diet*

[$0.76 per liter of diet, 1975]

Ingredient	Amount
Wheat germ	27 g
Nutrisoy	32 g
Sucrose	32 g
Wesson salt	9 g
Aureomycin	0.125 g
Choline chloride (10%)	9 ml
Vitamin B suspension[1]	1.5 ml
Ascorbic acid	3.6 g
Alphacel	4.5 g
Formaldehyde (10%)	3.3 ml
Methyl *p*-hydroxybenzoate	9 g
Water in blender	220 ml
Agar	22.5 g
Water for dissolving agar	550 ml
Potassium hydroxide (22.5%)	4.5 ml

[1] Same as that used by Berger (*2*).

The equipment used for mixing and dispensing the diets is shown in figure 2. The diets were dispensed into 30-ml plastic cups (10 ml of diet per cup) beneath a clean-air filter hood and allowed to gel (fig. 3).

In our second experiment, the following six diets were compared: (A) the soyflour and wheat germ diet of experiment 1; (B) a soyflour and wheat germ diet with agar reduced by 50% and substituted by 37 grams of corncob grits; (C) same as diet B with a 25% increase in soyflour; (D) the low agar, soybean, and wheat germ diet of experiment 1; (E) same as diet D plus 96 grams of sugar and 12 grams of wheat germ to equal the amount of wheat germ in diet A; (F) the same as diet E with 4.5 grams of alphacel added to equal the amount found in diet A.

Table 2.—*Composition of a pinto bean diet*

[$0.40 per liter of diet, 1975]

Ingredient	Amount
Wheat germ	40 g
Pinto beans	[1]173 g
Ascorbic acid	2.6 g
Formaldehyde (10%)	0.65 ml
Torula yeast (dried)	25.5 g
Methyl *p*-hydroxybenzoate	1.6 g
Sorbic acid	0.8 g
Water in blender	260 ml
Agar	10.2 g
Water for dissolving agar	251 ml

[1] 84 grams of beans soaked overnight and drained before use.

Table 3.—*Composition of a casein and wheat germ diet*

[$0.92 per liter of diet, 1975]

Ingredient	Amount
Wheat germ	27 g
Casein	27 g
Sucrose	45 g
Wesson salt	9 g
Aureomycin	0.25 g
Choline chloride	0.9 ml
Vitamin B suspension[1]	9 ml
Ascorbic acid	3.6 g
Formaldehyde (37.2%)	0.45 ml
Methyl *p*-hydroxybenzoate	1.35 g
Water in blender	229 ml
Agar	18 g
Water for dissolving agar	550 ml

[1] Same as that used by Adkisson et al. (*1*).

Ninety cups of diet were prepared from a single batch of each diet in experiment 1. Seventy-two cups of diet were prepared from a single batch of each diet in experiment 2.

TABLE 4.—*Composition of a low agar, soybean, and wheat germ diet*

[$0.32 per liter of diet, 1975]

Ingredient	Amount
Wheat germ	27 g
Nutrisoy	60.5 g
Sucrose	11 g
Wesson salt	9 g
Aureomycin	0.125 g
Choline chloride (15%)	6.25 ml
Vitamin B suspension	0.003 ml
Formaldehyde (10%)	3.75 ml
Methyl *p*-hydroxybenzoate	1.35 g
Sorbic acid	0.81 g
Water in blender	375 ml
Agar	3.75 g
Water for dissolving agar	375 ml
Corncob grits	62.5 g
Potassium hydroxide (4 *M* solution)	4.5 ml
Acetic acid (25%)	10 ml

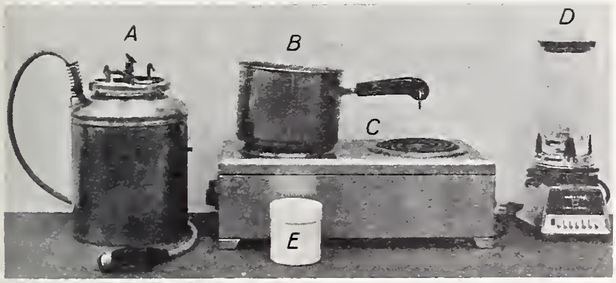

FIGURE 2.—Equipment used for mixing and dispensing the diets. *A*, Dispenser; *B*, pot for melting the agar; *C*, hot plate; *D*, blender for mixing diet; and *E*, carton containing diet ingredients.

FIGURE 3.—Clean-air filter hood under which diet is dispensed into cups and allowed to gel.

To obtain sugarcane borer larvae for the experiments, 25 pairs of adults were placed in a 1-gallon cardboard container lined with wax paper that served as the mating and oviposition surface. The moths were provided with a 15% sucrose and water solution, held at 26°±1° C, 90%±5% relative humidity, and kept on a 14 to 10 hour light to darkness (L:D) schedule. Wax paper linings were changed daily to collect the eggs. The eggs were disinfected for 3 minutes with a 3.5% formalin solution, followed by a thorough rinse in distilled water for 15 minutes.[4] Since the eggs do not separate or come off the wax paper during disinfection and rinsing, the papers and the eggs were air-dried beneath a clean-air filter. The dry papers and eggs were then placed into sterile 1-liter Ehrlenmeyer flasks containing moist cotton. The flasks were plugged with sterile cotton, covered with aluminum foil, and placed in a growth cabinet for egg hatch (30° C, 95% RH, and 14:10 L:D). Newly hatched larvae were immediately placed on the diet (one per cup). The cups were held at 26°±1° C and 50%±5% RH and kept on a 14:10 L:D cycle during the larval and pupal stages of development (figs. 4 and 5).

The times spent as larvae and as pupae were recorded in days, along with male and female pupal weights (in milligrams) on 2-day-old pupae. Pupation begins about 3 weeks after hatch, so the larvae were observed for 28 days to determine percentages of pupation, mortality, and nonpupation. Emergence begins about 1 week after pupation, so the pupae were observed for 14 days to determine percentages of emergence and mortality. The percentage of pupation represented the ratio of the number of pupated larvae (times 100) to the initial number of larvae, and the percentage of emergence was expressed as the ratio of the number of moths emerging (times 100) to the number of pupae. The number of deformed moths (incomplete emergence and failure to expand wings) was expressed as a percentage of the total number of emerged adults.

Nine growth characteristics were compared in both experiments: time as larvae, larval mortality, nonpupating larvae, pupation, time as pupae, pupal mortality, male and female pupal weights, emergence, and adult deformities.

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[4] E. G. King and G. G. Hartley, Bioenvironmental Insect Control Laboratory, personal communication, 1975.

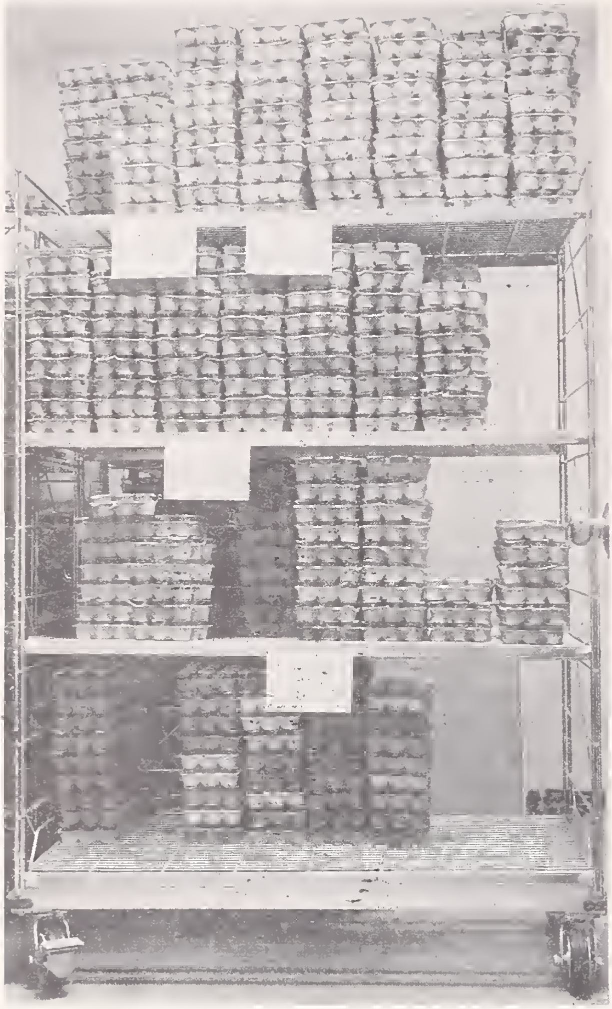

FIGURE 4.—Cups on storage cart within holding room.

FIGURE 5.—Cups with late-instar larvae (one per cup).

RESULTS

The data for experiment 1 are presented in table 5. The percentages of nonpupating larvae, pupation, pupal mortality, adult emergence, and adult deformities among the four diets were not significantly different ($P=0.05$). The larvae reared on the soyflour and wheat germ diet (A) had the shortest developmental period, and those reared on the pinto bean diet (B) had the longest. The percentage of larval mortality was the highest in larvae on the casein and wheat germ diet (C).

Development time as pupae was longer for larvae reared on the low agar, soybean, and wheat germ diet. The only significant difference observed in pupal weights was that male and female pupae produced on the pinto bean diet (B) and female pupae produced on the low agar, soybean, and wheat germ diet (D) were smaller.

The data for experiment 2 are presented in table 6. The percentages of pupal mortality and adult emergence were not significantly different ($P=0.05$). Larvae reared on diets B and C had the shortest developmental periods; those reared on diet D had the longest. Larval mortality was lowest on diet C and highest on diets D and E. Larvae grown on the other experimental diets did not vary significantly in mortality. The lowest percentage of nonpupating larvae was observed in larvae on diet A, and the highest percentage occurred in larvae on diet D. The highest percentage of pupation was observed in larvae reared on diet C and the lowest in larvae reared on diet D.

The pupal period was significantly shorter for pupae produced on diets B and C. There was no significant difference in pupal weight for female pupae produced on diets A, B, C, E, and F. Male pupae produced on diet F were significantly larger than those on the other diets except for diet E. The weights of male and female pupae produced on diet D was less than those on the other diets.

Adult deformities were significantly higher in larvae on diet D; no deformities were observed in moths produced on the other diets.

DISCUSSION

The soyflour and wheat germ diet appears to be the most desirable diet of those tested in experiment 1 for rearing the sugarcane borer. This

TABLE 5.—*Development of sugarcane borers on four artificial diets*[1]

Diet	Larvae					Pupae					
	Days in development	Number pupating	Mortality (%)	Nonpupating (%)	Pupation (%)	Days in development	Mortality (%)	Avg. wt. (mg)[2]		Emergence (%)	Adult deformities (%)
								Males	Females		
A[3]	20.8g	81	4.4f	4.6e	89.8e	7.7e	1.2e	103.6e (41)	176.8e (32)	100.0e	1.4e
B[4]	25.3e	83	3.6f	4.4e	92.0e	7.5e	2.2e	89.0f (43)	151.6f (33)	97.8e	1.2e
C[5]	21.9f	77	11.2e	3.4e	85.4e	7.7e	1.2e	104.2e (38)	188.2e (35)	98.6e	1.4e
D[6]	22.0f	79	9.6f	3.4e	87.4e	8.2f	2.4e	99.8e (37)	147.0f (30)	97.6e	1.4e

[1] Means of 90 replications. Means not followed by a common letter differ significantly at the 5% level as determined by Duncan's new multiple-range test.

[2] Figures in parentheses denote number of pupae used to obtain average weights.

[3] Soyflour and wheat germ.

[4] Pinto bean.

[5] Casein and wheat germ.

[6] Low agar, soybean, and wheat germ.

TABLE 6.—*Development of sugarcane borers on soybean and wheat germ diets*[1]

Diet	Larvae					Pupae					
	Days in development	Number pupating	Mortality (%)	Nonpupating (%)	Pupation (%)	Days in development	Mortality (%)	Avg. wt. (mg)[2]		Emergence (%)	Adult deformities (%)
								Males	Females		
A[3]	26.4ef	65	8.3ef	1.4f	90.3ef	8.7e	0.0e	92.8fg (31)	146.7ef (34)	100.0e	0.0e
B[4]	20.9h	63	12.5ef	8.5ef	87.5ef	6.8g	8.5e	95.0fg (28)	152.8e (29)	91.5e	0.0e
C[5]	21.4h	69	4.2f	7.2ef	95.8e	6.8g	7.2e	93.4fg (31)	143.0ef (33)	92.8e	0.0e
D[6]	27.2e	46	22.2e	13.9e	63.9g	8.9e	6.9e	86.6g (20)	114.2g (21)	93.0e	6.9f
E[7]	25.3efg	53	22.2e	4.2ef	73.6fg	8.7e	1.8e	102.6ef (24)	152.8e (27)	98.3e	0.0e
F[8]	24.9fg	59	12.5ef	5.6ef	81.9efg	8.7e	7.0e	109.9e (25)	145.7ef (25)	90.0e	0.0e

[1] Means of 72 replications. Means not followed by a common letter differ significantly at the 5% level as determined by Duncan's new multiple-range test.

[2] Figures in parentheses denote number of pupae used to obtain average weights.

[3] Soyflour and wheat germ.

[4] Soyflour and wheat germ with agar reduced by 50% and substituted by 37 grams of corncob grits.

[5] Diet B with a 25% increase in soyflour.

[6] Low agar, soybean, and wheat germ.

[7] Diet D plus 96 grams of sugar and 12 grams of wheat germ to equal the amount of wheat germ in diet A.

[8] Diet E with 4.5 grams of alphacel added to equal the amount in diet A.

diet costs about 30% less than the casein and wheat germ diet because soyflour is considerably cheaper than casein. The soyflour and wheat germ diet cost about 20% more than the pinto bean diet but was superior because of shorter larval development time, larger pupal weights, and ease of preparation (advance soaking or milling of materials not necessary).

At the Bioenvironmental Insect Control Laboratory, an excess of 1.5 million sugarcane borers were reared for at least 20 consecutive generations over the past 2 years (1973–75) on the soyflour and wheat germ diet without apparent deleterious effects. Over 1 million tachinid parasites [*Lixophaga diatraeae* (Townsend)] were reared during this same period on sugarcane borer larvae fed this diet. Larval survival on this diet averaged 92% (range 87%–96%) and time from oviposition to pupation averaged 28 days for this time period. These data were derived from records kept for brood colony progagation and large-scale host larval propagation for rearing *L. diatraeae.*

Because of the results of experiment 1, the second experiment was conducted to determine the combination of ingredients and amounts in the soyflour and wheat germ diet and low agar, soybean and wheat germ diet that would produce a suitable economical diet. Since the low agar, soybean and wheat germ diet costs about 32% less than the soyflour and wheat germ diet, we attempted to improve the quality of the low agar, soybean, and wheat germ diet for sugarcane borer development.

Results of experiment 2 indicated that the low agar, soybean, and wheat germ diet could be improved by additional sugar and wheat germ (the sugar content is probably the essential factor since there is little difference in the wheat germ content of this diet and that of the soyflour and wheat germ diet). The soyflour and wheat germ diet was improved, particularly in respect to shorter larval and pupal developmental periods, when the agar was reduced by 50% and corncob grits were substituted for the agar. Additional protein (soyflour) did not appear to improve the quality of the soyflour and wheat germ diet.

Further tests using more insects reared on diets from experiment 2 and possibly other modifications of these diets, for example, a new kind of agar-reducing material, will be continued in search of a more economical, yet nutritionally adequate, diet for rearing the sugarcane borer. Each major ingredient in these diets will be studied to determine how it affects particular growth characteristics. The cost of the diet will be considered before mass producing the borer. This work and related research will be oriented towards the goal of mass producing the sugarcane borer for consistent economical production needed in carrying out the biological control programs at this laboratory.

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